A Test of the Effectiveness of Permethrin to Reduce Arthropod Abundance and Increase Nesting Success of the House Wren (*Troglodytes aedon*)

Research Thesis

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By

Rebecca J Elting

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Project Advisor: Associate Professor Jacqueline K. Augustine, Department of Evolution, Ecology and Organismal Biology

Abstract

Parasites often induce stress in their host, impacting hosts' survival and reproductive success. This impact may be significant to the host, or hardly noticeable, depending on the parasite and intensity of the infestation. Some commensal organisms can be neutral or beneficial to the organism, such as when they feed on detritus. In birds, parasites not only appear on the organism itself but can reside in their nest. Although most birds show a negative correlation between parasite loads and nestling growth, house wrens appear to have a non-significant impact from parasite loads. One way to determine the effect of parasites is to fumigate the infested area with insecticides. This study has two purposes. First, to determine if arthropod infestation affected nesting success of house wrens (*Troglodytes aedon*). Secondly, this study looked at the distribution of arthropods in nests treated with permethrin or sprayed with water as a control. Previous studies on wrens did not examine permethrin as a fumigate. Nesting success and mite abundance data was collected from 72 first nests and 19 re-nesting attempts between April to August 2015 in Lima, Ohio. Nestlings were weighed 10 days after their hatch date. Arthropods were collected using a burlese funnel trap and counted from the nests after the nestlings had fledged. Overall, permethrin significantly had an effect on mites but not on ants. Mite and ant load had no effect on nestling success, even when parasite load was significantly reduced. House wrens seem to have behaviors (such as removing old nest materials) that allow them to keep mite infestations below harmful levels. Although we did not see a correlation between mite load and nesting success, numerous sublethal effects could be considered in the future. Further research should utilize a comparative framework to determine when mite infestations affect nesting success, because some species show negative correlation and others seem to be unaffected.

Introduction

Parasites often induce stress in their host, impacting hosts' survival and reproductive success (Martínez et al. 2011). This impact may be significant to the host, or hardly noticeable, depending on the parasite and intensity of the infestation. Some commensal organisms can actually be neutral or beneficial to the organism. For example, some species of non-parasitic mites feed on dermal detritus, or on other blood-sucking mites (Procter and Owens 2000).

Multiple species of parasites can feed on birds at any point in their life (Johnson and Albrecht 1993), but hosts are especially vulnerable during the reproductive season (Martínez et al. 2011). Parasites do not only appear on the organism itself, but often can reside in the nests of birds (Procter and Owens 2000). Parasites can cause heavy nestling mortality (Young 1993), and significantly decrease nestling weight (Merino et al. 2001). High blood-feeding mite loads are the cause for low hematocrit and small body size in pied flycatchers (*Ficedula hypoleuca*; Potti and Merino 1996, Potti et al. 1999), and low hatching success and post-fledging survival in rock doves (*Columba livia*; Clayton and Tompkins 1995) and barn swallows (*Hirundo rustica*; Møller 1990). Other studies have shown a correlation between high mite load and bright plumage (Proctor and Owens 2000).

Not all studies show a negative correlation between parasites and the birds' health. Some studies have shown no correlation between haematophagous mite load and sexual display performance (Proctor and Owens 2000). In red grouse (*Lagopus lagopus*), although female tick burden was experimentally reduced, there was no difference in either brood size at hatching, or brood size up to one month after hatching (Mougeot et al. 2008). Red-billed choughs (*Pyrrhocorax pyrrhocorax*) were in better condition with higher mite loads than those with lower mite loads (Blanco et al. 1997). Because mites can have either positive, negative or neutral effects, more studies are needed to determine when mites significantly affect reproductive success.

One way to determine the effect of parasites is to fumigate the infested area with insecticides. When nests of the mangrove finches (*Camarhynchus heliobates*) are sprayed with a 1% permethrin solution, no parasites were in the nests, and 95% of the experimental nests fledged offspring, compared to only 65% in the control group (Knutie et al. 2014). In another study, red-rumped swallow (*Hirundo daurica*) nests were fumigated with pyrethrin and piperonyl butoxide, and the experimental nests had higher nestling mass and survival (Merino et al. 2001). Fumigates such as permethrin show significant parasite mortality because of the chemicals' ability to interfere with the arthropods' sodium channels in neurons causing muscles to spasm, and eventually leading to paralysis and death (Toynton et al. 2009). Permethrin has a low toxicity in birds (Toynton et al. 2009).

Although most birds show a negative correlation between parasite loads and nestling growth, house wrens appear to have a non-significant impact of parasite load. In naturally occurring infestations of nests, house wren nestlings did not show a higher mortality or reduced growth of tarsi and primary feathers (Pacejka et al. 1996). When nests were heat-treated to kill ectoparasitic mites, house wren reproductive success was not affected (Johnson and Albrecht 1993). When nests fumigated with 10% Malathion solution were compared to non-fumigated nests, clutch size, nestling mass, and length of the nestling period showed no significant difference at different levels of mite population (Pacejka et al. 1998). Even mites with high transmission rates seem to not significantly affect house wrens (Pacejka et al. 1996).

My study has two purposes. First, I determined if arthropod infestation reduced nesting success of house wrens (*Troglodytes aedon*). Second, I looked at the distribution of arthropods in nests treated with permethrin or sprayed with water as a control. I predicted that the use of permethrin will decrease the arthropod population. Studies on birds suggests that mites reduce nestling size (Merino et al. 2001), however, studies on house wrens do not find the same effect (Johnson and Albrecht 1993; Pacejka et al. 1996, 1998). Previous studies on wrens did not examine permethrin as a fumigant.

Methods

Study site

The research was conducted between April and August, 2015. Data was collected from three locations (a woods, 40.7363927ºN, -84.0266254ºW; a golf course, 40.752005ºN, -84.036931ºW; and a park, 40.735647ºN, 84.029853ºW), with 40 nest boxes at each location. Every box had a width of 10.1 cm, a length of 14.0 cm, and a depth of 20.3 cm. The hole of box was 2.5 cm from the top of the box. Each hole had a diameter of 2.9 cm, which excludes many species of birds, except for house wrens.

Study Species

House wrens are small, insectivorous, migratory songbirds. They are about 11-13 cm long and have a mass between 10-12g (Johnson 2014). House wrens arrive on their breeding grounds around mid-April (Bent 1948). House wrens are often studied because of their preference for manmade bird houses, their tolerance for human activity, and their abundance in North America (Johnson 2014). Typically the females have a clutch size between 5-10 eggs, which incubate for 13 days with the average nestling period being 14-18 days (Johnson 2014). Nestlings usually reach their maximum mass by 12 days.

General Methods

We checked every nest box two times per week, until a nest cup was created. Afterwards, we checked the nest boxes daily to obtain the exact egg laying date. On the day the third egg was laid, the eggs were temporarily removed, and the nests were sprayed with 1% permethrin solution or distilled water. After allowing the nest to dry for 5 minutes, then the eggs were replaced. Because the average half-life of permethrin in aerobic soils is 39.5 days (Toynton et al. 2009), the permethrin should have remained effective throughout the nesting cycle (13 days for incubation plus 18 days for nesting period), but it should not have affected a second nest laid in the same box. After all the eggs are laid, nests were checked twice per week. At 13 days, daily nest checks resumed until 50% or more of the eggs hatched. After a hatch date was determined, nest boxes were checked every 3-4 days until the nestlings were 10 days old. When nestlings were 10 days old, they were weighed within 0.1 g and banded with a uniquely numbered leg band.

Five days after the nestlings are weighed and banded; nest activity was checked daily at a distance to prevent premature fledging. Once the parents stopped visiting the nest and there were no nestlings present, the nests were removed from the box and placed in a sealable plastic bag to transport it back to the lab to extract the mites. The mites were extracted using a burlese funnel trap, which causes the arthropods to move away from the hot light placed above the nest, towards a funnel placed below the nest, and down into a jar containing 70% ethanol. The sample was diluted to 50 ml, and mites present in three subsamples of 5 ml each were quantified. The mites were counted in all the boxes, regardless of whether they were treated or untreated.

Statistics

To test the effectiveness of permethrin, the number of arthropods in treated nests were compared to untreated nests. If the chemical was effective, the treated nests should have had a lower number of arthropods when compared to the untreated nests. To find out if the arthropod load affected nestling success, we compared the average mass of the nestlings within a clutch to the arthropod load of that nest. All analyses used nonparametric statistics because of the non-normal distribution of the number of arthropods. All analyses were conducted in JMP ver 11.0.0 (SAS Institute Inc. 2013). Sample size varied because it was not possible to measure every variable for every nest.

Results

Of the 120 boxes available, 72 boxes had at least one house wren nest which was used for mite extraction. There was a total of 91 nests collected; 72 first nests and 19 re-nesting attempts. Of the 91 nests, 89 nests had the mass recorded. The average mass of the nests was 80.02 ± 3.5g. The number of mites and ants found in nests showed a large variability but was generally low (Table 1).

Permethrin had an effect on mites but not on ants. First nests treated with permethrin had fewer mean mites than water-treated nests ($Z=5.69$; $N= 33$ control, 39 permethrin; $P< 0.0001$; Figure 1A). Second nests treated with permethrin tended to have fewer mean mites than water-treated nests ($Z=1.86$; $N= 7$ control, 12 permethrin; $P= 0.06$; Figure 1B). There was no effect of permethrin on the mean ants in the first nests (Z=-1.23; N= 33 control, 39 permethrin; P=0.22) (;Figure 1C) or in second nests (Z=0.81; N= 7 control, 12 permethrin; P= 0.42; Figure 1D).

Mite and ant load had no effect on nesting success. Clutch size did not differ between first nests treated with permethrin and water-treated nests (Z=0.98; N= 33 control, 36 permethrin; P= 0.33). Permethrin had no effect on mean nestling mass (Z=-0.49; N= 32 control, 39 permethrin; P=0.63) or mean nestling wing size (Z=1.31; N=32 control, 39 permethrin; P=0.19). No effect was found on clutch size in second nests treated with permethrin compared to water-treated nests (Z=-0.58; N= 7 control, 11 permethrin; P= 0.56). There was no effect of permethrin found in mean nestling mass (Z=0.34; N=7 control, 10 permethrin; P=0.73) or mean wing size (Z=0.05; N=7 control, 10 permethrin; P= 0.96) in second nests.

Discussion

Although permethrin did reduce mite populations when compared to water-treated nests, there was no correlation between our permethrin treatment and number of nestlings or nestling size or mass. Some studies found a correlation between high parasite load and decreased nesting success (Martínez et al. 2011, Merino et al. 2001, Møller 1991, Quiroga and Reboreda 2012 & Knutie et al. 2014). These studies were conducted on other birds, not house wrens specifically, which is a possible reason for differing conclusions. Other studies on house wrens found no significant correlation between nesting success and high parasite load (Pacejka et al. 1996, 1998; Johnson and Albrecht 1993, Johnson et al. 1991). What is remarkable is that no relationship between parasite load and nestling characteristics have been found in any house wren study, even if another parasite was considered (Johnson et al. 1991 and Johnson and Albrecht 1993 examined blow fly populations) or if other methods were used to eradicate parasites (heat treatment:

Pacejka et al. 1996, Johnson and Albrecht 1993; fumigation with 10% malathion solution: Pacejka et al. 1998). Both treatments did reduce ectoparasite population, similar to this study.

Although in this study, fumigating nests was chosen to eliminate ectoparasites, other methods like heat–treating the nest or using diatomaceous earth could have been utilized. Permethrin is an effective fumigate and is relatively non-toxic to vertebrates but is listed as a likely carcinogenic substance if ingested under the United States Environmental Protection Agency (Toynton et al. 2009). Permethrin is a general pesticide, but can be harmful to aquatic life and kill non-target organisms such as bees (Toynton et al. 2009). Unlike permethrin, diatomaceous earth and heat-treating nests have smaller environmental impacts (Dawson 2004, Hund et al. 2015). Diatomaceous earth is an organic way to eradicate northern fowl mites (Bennett et al. 2011) but is not efficacious when compared to other treatment methods (Dawson 2004). Heat treating nests involves transporting nests to the lab for treatment, which is not always optimal, but overall a very effective way to eradicate arthropods from nests (Johnson and Albrecht 1993, Hund et al. 2015). Ideally if the laboratory is close to the study site, heat treatment of nests seem to be the most effective with the least environmental impact. Diatomaceous earth could be used if there is fear of affecting non-target organisms or if the study site is near aquatic ecosystems.

House wrens seem to have behaviors that allow them to keep mite infestations below harmful levels during the duration of the nesting period. For instance, male house wrens remove old nest material which significantly reduces initial mite population (Pacejka and Thompson 1996). Since many mites remain in the nest post fledging (Szabó et al. 2008), the removal behavior can be extremely advantageous. Mite populations increase exponentially during the nesting period (Pacejka et al. 1996), but mites may leave nestlings close to fledging (Szabó et al. 2008). Mites are sensitive to temperature and moisture, preferring relatively high temperatures and high moisture (Weigmann and Kratz 1987). Pacejka et al. (1998) suggested that the removal of the old nesting material by the males could affect the microclimate in the nesting cavity creating an unfavorable environment for mites, thus keeping the mite population below harmful levels during the nesting period. This could explain why in our study, we did not find a correlation between nesting success and arthropod load. At the study sites, old nests are removed from nesting boxes each year and first nests were removed before second nests are initiated. The removal of the nest could have prevented mite populations from reaching lethal levels. However, Pacejka and Thompson (1996) found even when old nest were removed from boxes, it still did not significantly impact mite population levels. Pacejka et al. (1996) proposed that in addition to nest removal, the addition of spider egg cases could impact mite populations in nesting sites, allowing nestlings to fledge before mite populations reached virulent levels. Gable et al. (2019) found no significant reduction in mites when multiple spider egg cases were present in nests compared to those without spider egg cases. In general, the virulence of ectoparasites is often related to mode of transmission: vertically transmitted ectoparasites have a lesser effect on host fitness when compared to horizontal transmission (Clayton and Tompkins 1994). Even when ectoparasite loads were raised to unnaturally high levels, the effects seem to be benign if transmitted vertically (Tompkins et al. 1996). There is also evidence that virulence can change over time if parasites co-evolved with their hosts (Toft and Karter 1990).

Although we did not see a correlation between mite load and nesting success, numerous sublethal effects could be considered in the future. One thing our study lacked was the consideration of botfly load, which has been shown to be detrimental to house wren success

(Young 1993, Quiroga and Reboreda 2012). A huge factor in nestling success was the age at which nestlings are parasitized (Quiroga and Reboreda 2012). Our study did not record parasite load over the nesting season, but only recorded end parasite load. Another component our study did not consider was health of the individuals throughout the nesting season. A study done on house finches showed that conjunctivitis increased with mite load, but it did not negatively impact the birds (Davis 2013). While the birds in our study were not screened for illness, the lack of correlation between nesting success and mite load seem to follow the trend of similar studies. Also, many mites feed on detritus and oil, which accumulates during nestling growth, so the mites may benefit the host (Procter and Owens 2000). In house sparrows, there is no relationship between nesting success, body size, immunity and mite load (Szabó et al. 2008).

While we did find that permethrin reduced mite load, it did not significantly reduce ant load in house wren nests. Many studies show that permethrin is an effective fumigate against ectoparasites (Knutie et al. 2014, Toynton et al. 2009, Merino et al. 2001). Some studies showed permethrin was an effective way to control ants (Costa et al. 2005, Hara and Hata 1992), but it is not as effective against ants after 30 days (Rust et al. 1996). It is unclear why in our study, permethrin did not have a significant affect on decreasing ants and is an area of future study.

Further research should utilize a comparative framework to determine when mite infestations affect nesting success, because some species show negative correlation and others seem to be unaffected.

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Literature Cited

Bennett, D.C., Yee, A., Rhee, Y. J. and Cheng, K.M. (2011). “Effects of Diatomaceous Earth on Parasite Load, Egg Production, and Egg Quality of Free-Range Organic Laying Hens.” *Poultry Science* 90(7): 1416-1426.

Bent, A. C. (1948). “Life histories of North American Nuthatches, Wrens, Thrashers and Their Allies.” *Bulletin United States National Museum* 1-475.

Blanco, G., Tella, J. L. and Potti, J. (1997). “Feather Mites on Group-Living Red-Billed Choughs: A Non-Parasitic Interaction?” *Journal of Avian Biology* 28: 197-206.

Clayton, D. H. and Tompkins, D.M. (1994). “Ectoparasite Virulence is Linked to Mode of Transmission.” *Proceedings: Biological Sciences* 256(1347): 211-217.

Clayton, D.H. and Tompkins, D.M. (1995). “Comparative Effects of Mites and Lice on the Reproductive Success of Rock Doves (*Columba livia*).” *Parasitology* 110: 195–206.

Costa, H. S., Greenberg, L., Klotz, J. and Rust, M. K. (2005). “Response of Argentine Ants and Red Imported Fire Ants to Permethrin-Impregnated Plastic Strips: Foraging Rates, Colonization of Potted Soil, and Differential Mortality.” *Journal of Economic Entomology* 98(6): 2089-2094.

Dawson, R. (2004). “Efficacy of Diatomaceous Earth at Reducing Populations of Nest-Dwelling Ectoparasites in Tree Swallows.” *Journal of Field Ornithology* 75(3): 232-238.

Davis, A. and Cornelius, E. (2013). “Do Infections Lead to Higher Feather Mite Loads in Birds? A Test with Mycoplasmal Conjunctivitis in House Finches (*Haemorhous mexicanus*).” *The Auk* 130(4): 708-714.

Gable, C. L., Underwood, T. J. and Setliff G. P. (2019) "Do House Wrens (*Troglodytes aedon*) Add Spider Egg Cases to Their Nests for Heterospecific Cleaning?" *Canadian Journal of Zoology* 97: 50-56.

Hara, A. H. and Hata, T. Y. (1992). "Ant Control on Protea in Hawaii." *Scoentia Horticulturae* 51: 155-163.

Hund, A. K., Blair, J. T. and Hund, F. W. (2015). "A Review of Available Methods and Description of a New Method for Eliminating Ectoparasites From Bird Nests." *Journal of Field Ornithology* 86(3): 191-204.

Johnson, L.S. (2014). House Wren (*Troglodytes aedon*), The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology; Retrieved from the Birds of North America Online.

Johnson, L.S. and Albrecht, D.J. (1993). "Effects of Hematophagous Ectoparasites on Nestling House Wrens, *Troglodytes aedon*: Who Pays the Cost of Parasitism?" *Oikos* 66: 255-262.

Johnson, L.S., Eastman, M.D. and Kermott, L.H. (1991). "Effects of Ectoparasitism by Larvae of the Blow Fly *Protocalliphora-parorum* (Diptera, calliphoridae) on Nestling House Wrens, *Troglodytes-aedon*." *Canadian Journal of Zoology-Revue Canadienne de Zoologie* 69(6): 1441-1446.

Knutie, S.A., McNew, S.A., Bartlow, A.W., Vargas, D.A. and Clayton, D.H. (2014). "Darwin's Finches Combat Introduced Nest Parasites with Fumigated Cotton." *Current Biology* 24: R 355- R 356.

Martínez, J., Merino, S., Tomás, G., Moreno, J., Morales, J., Lobato, E. and Martínez, J. (2011). "Nest Ectoparasites Increase Physiological Stress in Breeding Birds: An Experiment." *Naturwissenschaften* 98: 99-106.

Merino, S., Martínez, J., Møller, A., Barbosa, A., Lope, F. and Rodríguez-Caabeiro, F. (2001). "Physiological and Haematological Consequences of a Novel Parasite on the Red-rumped Swallow *Hirundo daurica*." *International Journal for Parasitology* 31: 1187-1193.

Møller, A. P. (1990). "Effects of Parasitism by a Haematophagous Mite on Reproduction in the Barn Swallow." *Ecology* 71: 2345–235.

Møller, A. P. (1991). "Ectoparasite Loads Affect Optimal Clutch Size in Swallows." *Functional Ecology* 5(3): 351-359.

Mougeot, F., Moseley, M., Leckie, F., Martinez-Padilla, J., Miller, A., Pounds, M. and Irvine, J. (2008). "Reducing Tick Burdens on Chicks by Treating Breeding Female Grouse with Permethrin." *Journal of Wildlife Management* 72: 468-472.

Pacejka, A., Gratton C. and Thompson C. (1998). "Do Potentially Virulent Mites Affect House Wren *(Troglodytes aedon*) Reproductive Success?" *Ecology* 79: 1797-1806.

Pacejka, A., Santana, E., Harper R.G. and Thompson F. (1996). "House Wrens (*Troglodytes aedon*) and Nest-Dwelling Ectoparasites: Mite Population Growth and Feeding Patterns." *Journal of Avian Biology* 27: 273-278.

Pacejka, A. and Thompson, C. (1996). "Does Removal of Old Nests from Nestboxes by Researchers Affect Mite Populations in Subsequent Nests of House Wrens?" *Journal of Field Ornithology* 67(4): 558-564.

Potti, J., Moreno, J., Merino, S., Frías, O. and Rodríguez, R. (1999). "Environmental and Genetic Variation in the Haematocrit of Fledgling Pied Flycatchers *Ficedula hypoleuca*." *Oecologia* 120: 1-8.

Potti, J. and Merino, S. (1996). "Parasites and the Ontogeny of Sexual Size Dimorphism in a Passerine Bird." *Proceedings of the Royal Society. Biological Sciences* 236: 9-12.

Proctor, H. and Owens, I. (2000). "Mites and Birds: Diversity, Parasitism and Coevolution." *Trends on Ecology & Evolution* 15: 358-364.

Quiroga, M. A. and Reboreda, J. C. (2012). "Lethal and Sublethal Effects of Botfly (*Philornis seguyi*) Parasitism on House Wren." *The Condor* 114(1): 197-202.

Rust, M. K., Haagsma, K. and Reierson, D. A. (1996). "Barrier Sprays to Control Argentine Ants (Hymenoptera: Formicidae)." *Journal of Economic Entomology* 89(1): 134–137

Szabó, K., Szalmás, A., Liker, A. and Barta, Z. (2008). "Adaptive Host-Abandonment of Ectoparasites Before Fledging? Within-Brood Distribution of Nest Mites in House Sparrow Broods." *The Journal of Parasitology* 94(5): 1038-1043.

Toft, C. A. and Karter, A. J. (1990). "Parasite-host coevolution." *Trends in Ecology & Evolution* 5(10): 326-329.

Tompkins, D., Jones, T. and Clayton, D. (1996). "Effect of Vertically Transmitted Ectoparasites on the Reproductive Success of Swifts (*Apus apus*)." *Functional Ecology* 10(6): 733-740.

Toynton, K., Luukinen, B., Buhl, K. and Stone, D. (2009). *Permethrin Technical Fact Sheet*; National Pesticide Information Center, Oregon State University Extension Services.

Weigmann, G. and Kratz, W. (1987). "Oribatid Mites in Urban Zones of West Berlin Germany." *Biology and Fertility of Soils* 3(1-2): 81-84.

Young, B.E. (1993). "Effects of the Parasitic Botfly *Philornis carinatus* on Nestling House Wrens, *Troglodytes aedon*, in Costa Rica." *Oecologia* 93: 256-262.

Table 1: Distribution of mean mites and ants with standard error counted in three subsamples collected from 91 house wren nests from Allen County, OH, April- August 2015.

	Mean	SE	Range
Black Mites	2.5	0.78	0.0-63.3
White Mites	24.8	4.52	0.0-478.0
Red Mites	17.6	4.43	0.0-502.0
Ants	10.5	1.08	0.0-104.7

Figure 1. Permethrin caused the number of mites to decline in first nests (A) and second nests (B) but did not cause the number of ants to decline in first nests (C) and second nests (D) in house wrens in Lima, Ohio, April-August 2015.